Using Polyclonal and Monoclonal Antibodies in Regulatory Testing of Biological Products

Nancy E. Coe Clough and Paul J. Hauer

Abstract

Polyclonal and monoclonal antibodies are often used in regulatory testing of biologicals (vaccines and related products). One of the most common applications for antibody-based immunoassays is as a batch release test. Batch release tests, whether they measure serological responses to vaccination or they quantify individual antigens by in vitro methods, must provide an acceptable estimate of potency of an individual batch of vaccine. Thus, due consideration must be given to the type of antibody used or quantified in such assays. Differences in specificity and avidity may affect the utility of an assay as an indicator of potency; case examples are given to illustrate these concepts. Concerns associated with antigen quantification assays (e.g., reagent denaturation upon binding to solid substrates, and interference from nontarget antigens or additives in a complex vaccine) are also discussed. International efforts to harmonize test methods in recent years have increased the importance of establishing standardized antibodies. Sources of such antibodies and issues associated with the ongoing availability of antibody supplies are described.

Key Words: assay design; biologicals; monoclonal antibody; polyclonal antibody; vaccine potency; vaccine regulation

Introduction

Antibody-mediated procedures and assays are widely utilized by the biologics industry and regulatory authorities to evaluate and test biological products (e.g., vaccines and related products). One of the most critical applications from a regulatory standpoint is the batch release assay. For veterinary products, the efficacy of a vaccine is usually demonstrated in host animals during the prelicense (preregistration) phase of regulatory review. An acceptable surrogate, utilizing a laboratory animal model or in vitro technologies, is generally developed for routine batch testing (potency testing) of approved vaccines. The batch release assay must provide an acceptable assurance that each batch of product will be efficacious.

The potency of veterinary vaccines containing live microorganisms is usually evaluated by measuring the titer of the vaccine organism. Batch release tests for killed veterinary vaccines, however, are often antibody-mediated assays. Antigen quantification immunoassays, which use specific antibodies to detect individual antigens within complex vaccines, are increasingly popular in the United States. Such in vitro immunoassays are often used in lieu of animal-based testing. Assays that measure serological responses to vaccination, however, are also widely used, especially in the European Union.

There are myriad applications for antibodies in biological testing. It is outside the scope of this paper to provide a detailed review of all applications. This paper is intended instead to present broad concepts associated with antibody use in regulatory testing and to increase the reader's awareness of issues that must be considered when developing an assay for regulatory use. Case examples are included to illustrate selected concepts. The examples are drawn from our own laboratory experiences as veterinarians involved in the regulation of veterinary biologics in the United States, but similar situations may be encountered elsewhere.

Selecting an Antibody for a Specific Application

Designing and implementing an antibody-mediated assay for use in regulatory testing merits careful planning before entering the laboratory. A careful and thorough conceptualization of the assay can result in a more scientifically sound end product and a more efficient use of developmental resources. This conceptualization phase, however, is frequently ignored, or only superficially addressed, in the haste to move a biological product as quickly as possible toward licensure or registration.

Not all antibodies are created equal. This statement seems obvious enough, but it is all too easy to oversimplify the decisions associated with selecting the best antibody for a specific application. There is the temptation to use antibodies that we have in our laboratories or that are readily available, but the best antibody for the application may be

Nancy E. Coe Clough, D.V.M., Ph.D., is a Senior Staff Veterinarian, and Paul J. Hauer, D.V.M., M.S., Ph.D., is a Section Leader in Bacteriology Policy, Evaluation, and Licensing, the Center for Veterinary Biologics, Animal Plant Health Inspection Service, US Department of Agriculture, Ames, Iowa.

[1]Abbreviations used in this article: CVB, Center for Veterinary Biologics; ELISA, enzyme-linked immunosorbent assay; IU, international unit; IU/mL, international units per milliliter; MAb, monoclonal antibody; MAT, microagglutination test.

one that must be developed specifically for the purpose. Monoclonal antibody (MAb[1]) technology enables us to produce epitope-specific antibodies in most laboratory settings. Emerging antibody engineering technology (reviewed in Roque et al. 2004) may allow us even greater flexibility in the future.

Specificity

The integrity of any antibody-mediated potency test depends on the specificity of the antibody. We must be sure that we are evaluating clinically important antigens (or epitopes), with negligible cross-reactivity from related antigens. Historically, many assays measured antibody responses to whole vaccine organisms. We now know that serological titer is not well correlated with clinical protection against many diseases, and it is less frequently associated with protection to the degree necessary to make serological titer an adequate predictor of vaccine efficacy. Even for diseases in which serological titer is adequately correlated with clinical protection, the correlation is often based on specific antibodies directed against selected antigens within the organism (e.g., virulence factors). A serological potency assay should specifically measure that portion of the total antibody response that contributes to clinical immunity.

Likewise, in vitro antigen quantification assays should be carefully designed to provide the most useful information possible. When we use an in vitro assay, we lose the advantage of using an animal's immune system to look at the interactions among product components (e.g., immunomodulatory effects of adjuvant, competing effects among antigens in complex products). In vitro antigen quantification assays are used under the critical assumption that by evaluating one, or a few, key antigens in a complex product, it is possible to estimate the potency of the complete product. This assumption depends on a high degree of batch-to-batch consistency; thus, in vitro assays should be used only in conjunction with strict manufacturing controls.

The antibody in an in vitro immunoassay should quantify an antigen(s) that induces protective immunity in vaccinates. There are often several antigens in an organism that elicit protective immune responses, but there may be one, or a few, antigens that are the most relevant. Often the native three-dimensional conformation of an antigen is also critical to induce protective immunity; such is the case with many bacterial toxins. Particularly in the case of labile antigens, the assay should be able to discriminate between antigens that are in the proper conformation to be effective immunogens and those that may have been inappropriately denatured during vaccine manufacture.

Case Example: The Center for Veterinary Biologics (CVB[1]) has investigated possible alternatives to codified vaccination-challenge assays used to measure the potency of clostridial toxoids. The codified US potency test for *Clostridium sordellii* toxoid (Anonymous 2004) involves vaccinating rabbits, then harvesting immune serum. The serum is mixed with a defined amount of *C. sordellii* lethal toxin and administered by injection into mice. The neutralizing effect of the serum is quantified in international units. To avoid challenging mice with potentially lethal doses of toxin, an enzyme-linked immunosorbent assay (ELISA[1]) was developed to quantify the antitoxin antibodies in the rabbit serum. The ELISA was determined to be adequately sensitive and specific to detect rabbit antibodies directed against *C. sordellii* lethal toxin.

The ELISA was then compared with the codified in vivo (mouse) test, and the differences between the two approaches became apparent. *C. sordellii* toxoid preparations were experimentally prepared according to increasingly severe toxoiding protocols (Table 1; Hauer 1997). These preparations were used to inject groups of rabbits. The immune sera from each group of rabbits were tested by ELISA and the mouse test; the comparative results are shown in Figure 1.

The ELISA quantified the antibodies that bound to *C. sordellii* lethal toxin, but all antibodies, regardless of the epitope to which they bound, were measured. The mouse bioassay, however, effectively measured only that portion of the antibodies that could neutralize the toxin challenge. Antibodies that did not neutralize the toxin did not affect the outcome of the mouse assay. In this example, harsh toxoiding conditions quickly rendered the toxoid nonimmunogenic, but it was still antigenic. Thus, the total quantity of antibody generated by the rabbits was "satisfactory" in all treatment groups. The quality of the antibody response, however, was dramatically affected. As a predictor of vaccine efficacy, and thus as a batch release potency assay, the ELISA was unacceptable because it did not discriminate between protective and

Table 1 Increasingly severe protocols used to prepare toxoids to evaluate the ability of a new potency test to evaluate toxoid quality

Antigen content	Toxoiding protocol
1×	0.6% Formaldehyde; 37°C, 2 days
3×	0.9% Formaldehyde; 45°C, 1 hr; then 37°C, 2 days
6×	1.2% Formaldehyde; 50°C, 1 hr; then 37°C, 2 days
9×	1.6% Formaldehyde; 55°C, 1 hr; then 37°C, 2 days
12×	Boiled - 1 minute

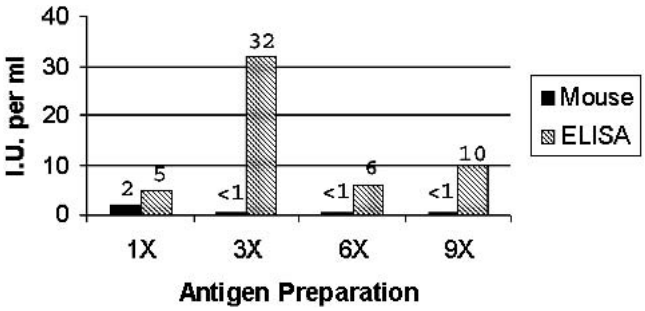

Figure 1 Comparison of a mouse bioassay (MOUSE) and enzyme-linked immunosorbent assay (ELISA) to measure the antitoxin titer of postvaccination serum of rabbits vaccinated with *Clostridium sordellii* toxoid. Toxoids were prepared according to increasingly severe conditions (see Table 1). Potency of the toxoid is considered satisfactory if the antitoxin titer is at least 1 international unit (IU) per milliliter.

nonprotective antibodies. By quantifying antibodies without adequate consideration for epitope specificity, the ELISA gave the false illusion of satisfactory toxoid potency when it did not, in fact, exist.

Taking a different approach, an antigen quantification assay using a toxin neutralizing MAb, is being investigated for *Clostridium hemolyticum* toxoid. The CVB developed a panel of MAbs against the beta toxin of *C. hemolyticum*. The beta toxin has been shown to be a key immunogen of *C. hemolyticum* (Hauer et al. 2004; Hauer, unpublished data). Toxin-neutralizing MAbs were identified for further study. Surface plasmon resonance (Van Regenmortel 2000) was used in preliminary studies, and the studies suggest that one of the candidate MAbs, which binds immunogenic toxin well, does not bind to denatured beta toxin (Yeary and Hauer, unpublished data). Thus, it may be possible to develop an ELISA that through the proper use of a biologically relevant antibody will quantify the amount of properly configured toxoid in biological preparations.

Affinity/Avidity

The affinity of an antibody preparation is also an important consideration. Affinity refers to the strength of the bond between antigen and antibody (reviewed in Harlow and Lane 1999). Antigen-antibody binding is an equilibrium reaction; binding and dissociation continually occur. High-affinity antibodies, however, tend to bind more quickly and to dissociate more slowly than low-affinity antibodies. High-affinity antibodies also tend to bind a larger proportion of the available antigen.

Functionally the "strength" of an antibody in a particular application depends on the avidity of the antigen-antibody complex. Avidity refers to the overall stability of the complex. It is affected not only by the affinity of the antibody but also by the valency of binding and the three-dimensional features of the complex. Those interactions that are stabilized by multivalent interactions and a tight three-dimensional structure tend to be stronger than those that are not.

In certain applications we may seek a high-affinity antibody. High-affinity polyclonal antibodies work well for antigen capture in sandwich ELISAs and in vitro neutralization assays. In other applications, however, an antibody with excessive affinity may be detrimental. Monoclonal antibodies are often desirable for competitive immunoassays that target precise epitopes, but if the affinity of the competing antibody is too high, very little competition may occur.

Case Example: Monoclonal antibodies were being evaluated for use in an ELISA to measure the potency of *Escherichia coli* bacterins. Although both MAbs bound to the same (or an overlapping) epitope, one had a much higher binding affinity than the other.

Serial dilutions of K88 *E. coli* pilus antigen were immobilized on an ELISA plate. Various MAbs, including the two candidate MAbs specific for K88, were added to individual wells of the plate and allowed to bind to the captured pilus antigen. A second MAb, which was enzyme labeled, was sequentially added to the plate as a competing antibody. The amount of bound labeled antibody was measured. As illustrated in Figure 2A, the high-affinity enzyme-labeled antibody (MAb #2) displaced essentially all of the unlabeled lower affinity antibody (MAb #1), making it appear as though the two antibodies did not compete for binding sites (i.e., the binding of labeled MAb #2 in the presence of unlabeled MAb #1 was similar to the negative control baseline). When the antibody configuration in the assay was reversed (Figure 2B), the ability of the two antibodies to compete for binding sites was apparent (i.e., the binding of labeled MAb #1 was substantially reduced from baseline in the presence of unlabeled MAb #2).

Impact of Assay Type or Format

An antibody that is ideal for one assay format may not work well in another. Sometimes it may merely reduce the efficiency of an assay, but in other situations, the antibody may not function at all in the alternate assay format.

Solid-phase assays rely on a support medium (e.g., plastic) to immobilize reagents. Antigens and antibodies alike may be denatured upon binding to the solid phase, altering their ability to bind their complementary ligand. Adsorption (binding) to solid supports relies primarily on hydrophobic interactions between the molecule and the support. Hydro-

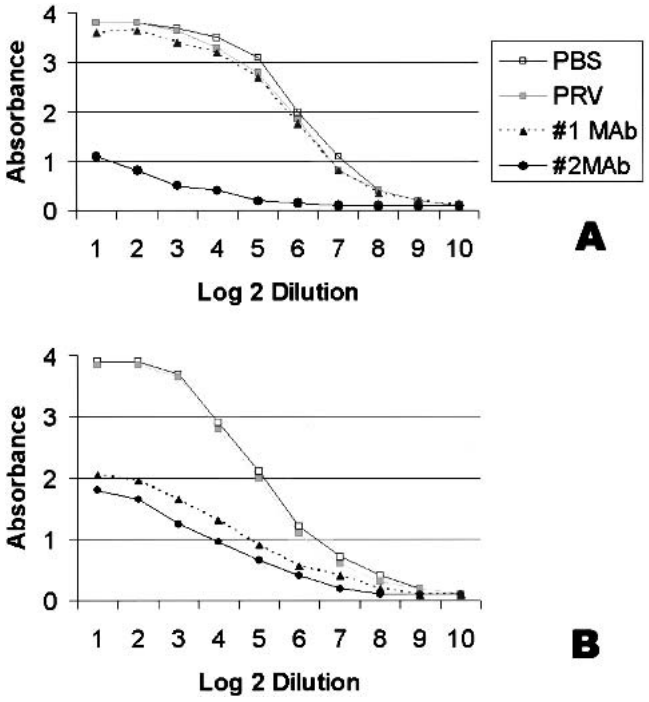

Figure 2 Competitive enzyme-linked immunosorbent assay to quantify *Escherichia coli* K88 pilus antigen. Various unlabeled antibodies were first added to plates containing bound antigen, as described on page 302 for Figure 2A. MAbs #1 and #2 were directed against K88 antigen, whereas a MAb against pseudorabies virus (PRV) and phosphate-buffered saline (PBS) served as negative controls. After incubating with the unlabeled antibodies, an enzyme-labeled detection antibody was added. In Figure 2A, a labeled preparation of MAb #2 was added. In Figure 2B, a labeled preparation of MAb #1 was added. The amount of bound, labeled antibody was determined.

phobic bonds rely on extremely close contact between the molecule and its substrate. Orientation is random. Highly flexible molecules may be more severely distorted than more rigid ones. When an antigen is structurally distorted upon binding to a solid substrate, antibodies recognizing conformational epitopes of that antigen may be more affected than those recognizing linear epitopes.

Case Example: Antiserum against *Leptospira pomona* was needed to use as a polyclonal capture antibody in a sandwich ELISA to measure the potency of mixed bacterins containing *L. pomona*. Rabbit antiserum was initially screened by the supplier in a microagglutination test (MAT[1]). The MAT titer against *L. pomona* was ≥1:102,400. The titer against *Leptospira grippotyphosa* was 1:200. On the basis of the MAT assay, the serum appeared highly specific for *L. pomona*. Although it would have been preferable to use an antiserum that was considered to have baseline reactivity against *L. grippotyphosa* (<1:100 titer), the extreme difference in titers by MAT suggested that it might be possible to dilute the serum to the extent that cross-reactivity to *L. grippotyphosa* would be negligible in the *L. pomona* ELISA. As hown in Table 2, the cross-reactivity was persistent, even at high serum dilutions, in the ELISA format. Thus, the serum, when used in the ELISA, was not nearly as monospecific as the MAT results would suggest.

Some assays (e.g., ELISAs) require enzyme-labeled detection antibodies. The primary detection antibody may be labeled directly, or an unlabeled detection antibody may be followed by an enzyme-labeled antispecies antibody that will bind to the unlabeled antibody. Enzyme-labeled antispecies antibodies, which are commercially available, are commonly used, but directly labeling the primary detection antibody may be desirable for some assays.

Direct labeling of the detection antibody eliminates the need for a second incubation step to add an antispecies conjugate, and it often reduces the background absorbance observed with some secondary conjugates. Not all antibody preparations, however, can be successfully labeled. The enzyme binds to specific ligands on the antibody molecule (e.g., biotin binds to free amino groups). If the necessary ligand is in the antigen-binding site of the antibody, the binding capacity of the antibody may be destroyed. This effect is more pronounced with MAb preparations because of their homogenous nature. Enzymes can bind at the same location on every antibody molecule in a MAb preparation, thus rendering the entire antibody population unusable. Conversely, it is likely that only certain molecules in the

Table 2 Relative ability of an antiserum, with a microagglutination titer of 1:102,400, and 1:200 to *Leptospira pomona* (LP) and *Leptospira grippotyphosa* (LG), respectively, to capture antigen in a sandwich enzyme-linked immunosorbent assay[a]

	Antigen dilution		
	1:4	1:8	1:16
Leptospira grippotyphosa	0.959	0.425	0.195
Leptospira pomona	1.313	0.637	0.287
S/N ratio	1.4	1.5	1.5

[a]The antiserum was diluted to 1:100,000 and added to the entire test plate. Serial dilutions of LP antigen were added to half of the plate, and LG antigen was added similarly to the other half. Monoclonal antibodies specific for the antigen were used to detect antigen bound by the capture antiserum. The data show the average absorbance value of replicate wells for each antigen dilution. Signal:noise (S/N) ratios were calculated for each data point. Negative control wells, containing no antigen, had an average absorbance of 0.004.

heterogenous antibody mixture in polyclonal antiserum would be affected.

Labeled antibodies also may not bind antigen efficiently (especially antigen bound to a solid substrate) because of steric hindrance. The various enzymes used in ELISAs have different molecular weights. Alkaline phosphatase, for example, is quite large in proportion to an immunoglobulin G antibody molecule (80,000 vs. 150,000 MW, respectively); it is more likely to cause steric hindrance than biotin (250 MW). Thus, consideration must be given not only to the antibody preparation but also to the choice of enzyme.

The same assay may be used to test a variety of different biological products. Assays also may be used to test manufacturing intermediates as well as completed product, which tends to be more complex and may contain interfering substances such as adjuvant. Antibodies that work well to test nonadjuvanted manufacturing intermediates may not bind adequately to antigen in completed product. Several treatments may be utilized to elute the antigen from its adjuvant before the sample is assayed, but elution procedures tend to be highly inefficient and free only a small proportion of the total antigen in the sample.

Standardization and Validation of Antibodies

Assay-to-assay repeatability is a critical feature of both initial assay validation and ongoing assay monitoring. This process requires, among other things, a standardized source of antibody. Adequate consideration should be given during assay conceptualization to antibody source and availability. We advise developing a plan to transition from one lot of antibody to the next to avoid disrupting the integrity of the assay.

World Health Organization (WHO[1]) Standards

In an attempt to standardize vaccine evaluation and diagnostic testing, the WHO has developed international standard antisera against many human and animal pathogens. (A current list of available standards is posted at www.who.int/vaccines/Biologicals/Ani1.asp.) Many of these standards are produced from high-titered equine serum. Standardized antisera that neutralize bacterial toxins make up a large percentage of the available standards. The antitoxins are assigned international units (IUs[1]) that are based on their neutralizing capacity, and IUs are used as the relative unit of comparison.

Many of the international standard antitoxins were developed before 1970, using relatively unpurified antigen. Many antibodies in these preparations react to bacterial proteins other than toxin, making them unattractive candidates for toxin-specific applications. Future antitoxin preparations may be improved by producing polyclonal antiserum that is more toxin specific, or by using a mixture of high-affinity toxin-neutralizing MAbs.

Other Standard Sources

Commercial sources of specific polyclonal antibodies or MAbs, genetically engineered antibodies, and nearly any other conceivable form of antibody are readily available. Standardized reagents for regulatory testing, however, are more difficult to obtain. Some non-WHO sources of national and international reference antibody preparations that are used in regulatory testing are listed in Table 3.

Transitioning Between Successive Lots of Antibodies

Changing the lot of any antibody in an assay, whether it is an integral part of the assay architecture or an external control preparation, has the potential to change assay performance and adversely affect comparisons of results from different assay runs. Thus, each new lot of antibody must be appropriately validated against a suitable standard before it is used in official testing. The standard may be an internationally recognized serum lot, or a new lot of antibody may be compared with the lot immediately preceding it. If a new

Table 3 Sources of international and national reference antibodies

Organization	Website
World Organization for Animal Health (OIE)	www.oie.int/eng/normes/en_sera.htm
European Directorate for the Quality of Medicines	www.pheur.org/site/page_dynamique.php3?lien=M&lien_page=18&id=6
National Institute for Biological Standards and Control References (UK)	www.nibsc.ac.uk/
U.S. Department of Agriculture—National Veterinary Services Laboratories	www.aphis.usda.gov/vs/nvsl/reagentsfaddl.htm
US Department of Agriculture—Center for Veterinary Biologics	www.aphis.usda.gov/vs/cvb/memos/memo800_97.pdf

lot is compared with the lot immediately preceding it, however, consideration must be given to the potential that negligible differences between individual lots may accumulate over multiple generations and have significant effects over time. Fortunately antibody preparations (especially antiserum) tend to be highly stable when frozen under controlled conditions, so it is often possible to validate against a single standard for many years.

An advantage of using MAbs is that provided the hybridoma cells are stably secreting immunoglobulin, each lot of MAb should be identical in affinity and specificity. Supplies of consistent MAb are theoretically limitless. This abundance, however, does not imply that it is unnecessary to validate each new lot of MAb carefully.

The transition from one lot of polyclonal antibody to another is more complicated. Even though they may be purified for isotype and a certain degree of antigenic specificity, polyclonal antibodies remain a complex mixture of individual molecules that bind with differing affinities to different antigenic epitopes. It is impossible to match two lots of polyclonal antibodies to the degree achievable with MAbs, but attempts should be made to match properties as closely as possible and to understand the limitations of comparing results of assays performed with different lots of antibody.

Case Example: Because international standards are available only in limited quantities, it is common procedure to produce a secondary standard, matched to the international standard, for routine use. The neutralizing capacity of serum, however, depends both on the quantity of antibody molecules (i.e., the proportion having the correct epitope specificity) and on the avidity with which they bind to their target antigen. The need to select an "avid" serum for an antitoxin preparation is well documented (Batty 1971). New assay methods demonstrate, however, that avidity can vary widely even among individual lots of hyperimmune sera that would have been previously classified identically as avid.

Antitoxin preparations having the same neutralizing capacity (international units per milliliter [IU/mL[1]]) can vary qualitatively, as demonstrated in Table 4. Six tetanus antitoxins were prepared. All of the preparations were closely matched in terms of IUs/mL, as determined by mouse assay. When tested by competitive ELISA against a low-affinity competing MAb, however, the preparations had different apparent potencies (Kolbe and Hauer, unpublished data). Further investigation suggested that differing average affinities may in part account for the observed differences.

Thus, one should not assume that secondary standards are equivalent to an international standard in all respects. In general, secondary standards should be used only at dilutions for which direct comparisons with the international standard have been established. For example, an international standard containing 10 IU/mL can, by definition, be diluted 1:10 to obtain a preparation containing 1 IU/mL. Due to differences in average affinity, a secondary standard when diluted by the same factor may not have a bioactivity of exactly 1 IU/mL. If a 1 IU preparation is needed, it is more appropriate to dilute the international standard to 1 IU and match the secondary standard to the diluted international standard. Otherwise, "unit drift" may occur. The same unit drift problem can be encountered when international standards are replaced when adequate attention is not given to matching the average affinities of the existing and replacement standards.

Table 4 Comparison of five tetanus antitoxin preparations (A-E)[a]

	A	B	C	D	E
Relative antibody concentration	1.0	1.6	1.3	1.8	1.9

[a]All of the preparations were standardized to 500 IU/mL by mouse bioassay. The relative antibody concentration reflects the relative absorbance (compared with preparation A) of each preparation at a 1:5000 dilution in an enzyme-linked immunosorbent assay. This single point was derived from the linear portion of the dose-response curves for each preparation and is representative of the measured relative potencies of the preparations.

Availability of Antibodies for Harmonized Use

When developing antibodies and antibody assays for use by multiple parties, one must give adequate consideration to the availability of the antibodies. They must be readily available in sufficient quantities to meet demand. As global interaction increases and regulatory requirements for testing biological products become more harmonized, the issues of maintaining adequate inventories for standard reagents and easy availability become even more critical.

Distribution by Developer

The developers of antibodies may elect to distribute the antibodies to requestors directly. The advantage of this method is that the developer maintains tight control over who receives antibodies. In the case of MAbs, the developer also may elect to provide secreted antibodies (ascites or bioreactor fluids) while restricting distribution of the hybridoma. The disadvantage of this method is that the developer must be confident that he or she has adequate resources and personnel to ensure a steady supply of high-quality

reagents because any lapse in supply could have critical repercussions for the industry that depends on the antibody.

Central Repositories

There are several not-for-profit organizations, including the American Type Culture Collection in the United States (www.atcc.org) and the European Collection of Animal Cell Cultures (www.ecacc.org.uk) in the United Kingdom. These organizations provide secure repositories for hybridomas. Depositors can elect to include their hybridoma in the organization's catalog. For a small fee, requestors may obtain hybridoma cells so that they may produce their own stocks of MAbs. Current procedures and policies for depositing hybridomas with these organizations are described on their websites.

Antibody Patents

Some inventors/developers elect to patent the MAbs they develop. In return for publishing information about the antibody, the applicant is granted exclusive rights over its use and distribution for a specified period of time. Patents are usually sought for antibodies with commercial application. More complete discussions of antibody patents are found elsewhere (e.g., Crawley 1995). Within the biologics arena, incentives may exist to patent antibodies used in commercially marketed diagnostic test kits. When developing standard antibodies to test the potency of biologics, however, there may be little value in patenting. Patenting may, in fact, provide a disincentive to regulatory authorities who are evaluating potential antibodies for standardized assays because distribution of such antibodies must be unrestricted.

Future Direction

Advances in molecular biology and genetic engineering provide almost limitless potential for the utility of antibodies. Contemporary techniques also provide opportunities to eliminate the use of animals in antibody development and production. Unfortunately, these advances are only very slowly becoming evident in the arena of regulatory testing. Most current national and international antibody references are not suitable for use in more advanced test methods.

It is incumbent upon regulators to utilize current technologies to create a new generation of antibody standards. Powerful new protein purification techniques could be used to prepare immunizing antigens that would generate highly specific conventional polyclonal antibody references. The use of pooled MAbs also should be investigated as a potential method to prepare consistent, standardized polyclonal preparations. The amount of time required to adopt an international standard should be decreased to allow quicker access to a standard prior to adopting various other test methods.

International collaboration to share reagents would facilitate the harmonization of regulatory test methods more than any other single effort. The problems involved with establishing a pool of standardized reagents appear to be much more of a political and economic issue than a technical problem. Scientific meetings have been held in recent years to facilitate collaboration in test development and reagent production, and proposals have been made for internationally recognized collections of standardized reagents (Hauer and Clough 1999; NIAID 2003). We recommend expansion of such efforts.

Summary

Antibodies play a critical role in the regulatory testing of biological products, especially in assays to ensure adequate potency of individual product batches. It is essential to exercise care when selecting an antibody for a potency assay because the specificity and avidity of the antibody can affect the suitability of the assay to predict potency with acceptable accuracy and precision. As our society becomes increasingly global, incentives increase to establish internationally recognized sources of standardized antibodies and to develop harmonized methods for testing biologicals. We encourage regulators to continue to expand their efforts toward collaboration on assay and reagent development.

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